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# (54) MODIFIED WHEY PROTEIN AND PROCESS FOR PRODUCING THE SAME

A modified whey protein having improved keeping qualities and comprising a partly heat-denatured whey protein and a casein protein optionally together with an undenatured whey protein. The partly heat-denatured whey protein can be prepared by heating a whey protein, the protein concentration and pH value of which have been adjusted respectively to 0.5 to 15 % by weight and 6 to 8, at 55 to 120 °C for 1 to 120 minutes. The process for producing the modified protein comprises adding a casein protein to the partly heatdenatured whey protein or adding a casein protein to an undenatured whey protein to partly denature the same by heating. The modified whey protein having improved keeping qualities can provide a gel which has a good water retention and a high elasticity and is useful as a food material.

#### Description

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#### FIELD OF THE INVENTION

The present invention relates to a processed whey protein and a process for manufacturing the processed whey protein. The processed whey protein of the present invention is useful as a food material for manufacturing foods requiring such properties as gelling capability, water retention capacity, and high viscosity.

#### BACKGROUND OF THE INVENTION

Whey proteins have conventionally been used as foodstuffs because of the high nutrition value possessed by the whey proteins themselves, and also have widely been used as supplemental materials for foods such as binders, extenders, and water retention agents, wherein the properties of the whey protein such as emulsifying property, foaming property, and gelation are utilized. Among these, the high gelation makes the whey protein an advantageous texture modifier for raw meat and fish meat products. Thus, the gelation of whey proteins is one of the important factor that improve the texture and water retention capacity of foods. Generally, proteins are denatured by heat and create mutual interactions such as hydrophobic interaction between the protein molecules, non-covalent bonds such as ionic bonds and hydrogen bonds and an SH/SS exchange reaction, whereas whey proteins are denatured and gelated by heat at temperatures of 60°C or higher. However, because the whey protein gel thus obtained is generally non-transparent and has only a small water retention capacity and a brittle structure, such a whey protein gel is not preferably used as a food material.

For these reasons, in order to improve the gel structure of whey proteins a partially heat-denatured whey protein solution produced by partially denaturing the whey protein by heating, or a solution obtained by drying this partially heat-denatured whey protein solution to produce a dried powder and re-dissolving the dried powder, is used to obtain a whey protein gel which has high water-retention capacity and excellent texture. Specifically, a highly elastic gel with a high water retention capacity can be obtained by producing a solution of the partially heat-denatured whey protein by heating a whey protein solution at a concentration of 4-15% by weight, preferably 5-12% by weight and by at 55°C to 120°C, preferably 65°C to 95 °C, or producing a solution by drying this solution and re-dissolving the resulting dried powder, and then by adding a salt at low temperatures to these solutions (Japanese published unexamined patent application No. 124067/1990), or by freeze-drying and thawing these solution (Japanese published unexamined patent applications No. 280834/1991 and 27249/1991).

Whey proteins which are usually spherical produce a soluble aggregate in which protein molecules are aggregated like chains, if partially denatured by heating. This soluble aggregate in which protein molecules of the whey protein are aggregated like chains are hereinafter referred to as "soluble aggregate", and the whey protein which contains this soluble aggregate is referred to "partially heat-denatured whey protein". Although the partially heat-denatured whey protein does not gel in the soluble aggregate state, this soluble aggregate of whey protein forms a three dimensional network and produces irreversible gel if the above-mentioned measures are taken. The whey protein gel thus obtained has a high water-retention capacity, superior elasticity, and a smooth constitution.

The gel produced from the solution of the partially heat-denatured whey protein which has been obtained by partially denaturing whey protein with heat has a good constitution. However, a solution of the partially heat-denatured whey protein tends to change its form into a sol-like material due to an increase in its viscosity during storage for a long period of time, because the whey protein which has been partially denatured and are present as a soluble aggregate readily reacts with a small amount of a salt in the solution.

# DISCLOSURE OF THE INVENTION

The inventors of the present invention have undertaken extensive studies to improve the shelf life of the solution of the above-mentioned partially heat-denatured whey protein. As a result, the inventors have found that if a casein protein is added to a solution of a partially heat-denatured whey protein wherein whey proteins are in the form of a soluble aggregate, the shelf life of the partially heat-denatured whey protein solution is improved, and a gel obtained from this whey protein exhibits a high water-retention capacity, excellent elasticity, and smooth constitution even if the the solution of the pertially heat-denatured whey protein is stored for a long period of time. These findings have led to the completion of the present invention. In addition, a powder obtained by drying this solution also has an excellent shelf life, and a gel obtained by dissolving this powder exhibits a high water-retention capacity, excellent elasticity, and smooth constitution. Accordingly, an object of the present invention is to provide a processed whey protein with improved shelf-life, which is capable of producing a gel exhibiting, a high water-retention capacity, excellent elasticity, and smooth constitution, and to provide a process for manufacturing such a processed whey protein.

The present invention has been achieved to solve these problems and relates to a processed whey protein with

improved shelf life comprising a partially heat-denatured protein and a case in protein and to provide a process for manufacting the same. This processed whey protein can be prepared by adding a case in protein to a solution of a non-denatured whey protein and heating the solution to partially heat-denature whey protein, or by producing a partially heat-denatured whey protein by heating a solution of a non-denatured whey protein and adding a case in protein to this partially heat-denatured whey protein solution.

In the present invention, it is possible to add a non-denatured whey protein together with a casein protein.

Furthermore, the solution of the processed whey protein may be dried and made into a powder, and the powder may be molded into any optional shape.

More specifically, the processed whey protein of the present invention can be obtained as a solution by a process comprising preparing a whey protein solution with a concentration of 15% or lower, a concentration range wherein the whey protein does not aggregate when heated at a temperature higher than the heat-denaturing temperature of the whey protein, i.e. 55°C or higher, adding a casein protein to this solution, and heating the resulting solution at a heat denaturing temperature of the whey protein, i.e. at 55-120°C. Alternatively, it is possible to prepare the whey protein solution with a concentration of 15% or lower, a concentration range wherein the whey protein does not aggregate when heated at a temperature higher than the heat-denaturing temperature of the whey protein, i.e. 55°C or higher, heat this solution at a heat denaturing temperature of the whey protein, i.e. at 55-120°C, and then add a casein protein to the resulting solution. The solution of the processed whey protein may be dried and made into a powder by a conventional method, and the powder may be molded with any optional means.

A whey protein solution is commonly known to aggregate by heating to produce a brittle gel, when the concentration is higher than 15%. To prevent such a gel formation, a solution of whey protein at a concentration of 15% or less, more preferably 12% or less, is heated to a temperature of 55 to 120°C, and more preferably to 65 to 95°C. This treatment causes the whey protein to be partially heat-denatured and causes a hydrophobic part to appear on the surface of the whey protein molecules which are spherical. Although this whey protein solution changes its properties depending on the degree of hydrohobocity of the whey protein, the target partially heat-denatured whey protein solution wherein the whey protein has been denatured to a soluble aggregate can be obtained by heating the solution with a concentration mentioned above and adjusted to pH 6-8 for 1 to 120 minutes. This partially heat-denatured whey protein solution can be prepared according to the method disclosed in Japanese published unexamined patent application No. 64550/1993.

When a whey protein solution is heated at a concentration which does not cause the whey protein to aggregate, the whey protein reaches a certain denatured state, whereby the SH/SS exchange reaction occurs. At the same time, its hydrophobic characteristics are increased. As a result, the molecules of whey protein are inter-associated and form a soluble aggregate. A feature of the present invention resides in improving the shelf life of partially heat-denatured whey protein by adding a casein protein to a solution containing this soluble aggregate of whey protein, i.e. a solution of partially heat-denatured whey protein.

The processed whey protein solution in the present invention is prepared as follows. First, a non-denatured whey protein aqueous solution with a whey protein concentration of 0.5 to 15%, preferably 5 to 12%, is prepared. If the concentration of whey protein in the non-denatured whey protein solution is less than 0.5%, the gelation is only insufficiently achieved when the gel is produced from the resulting processed whey protein. If the concentration of non-denatured whey protein is more than 15%, the solution becomes highly viscus during heating, so that a part or all of the whey protein form a gel.

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The non-denatured whey protein solution is adjusted to pH 6-8. If the pH is less than 6, the whey protein aggregates and precipitates during heating, so that whey protein gel with smooth constitution is formed only with difficulty. If the pH is more than 8, the taste of the whey protein gel obtained is poor. Moreover, it is desirable that the whey protein solution contains a salt-derived ion at a concentration of 0.5% or less, preferably 0.3% or less, and particularly preferably 0.01% or less. If the concentration of the salt-derived ion is more than 0.5%, the whey protein aggregates and precipitates, or gels during heating.

The processed whey protein of the present invention can be obtained by adding a casein protein to the non-denatured whey protein solution thus prepared and heating the solution to partially denature the whey protein, or by heating the non-denatured whey protein solution prepared above to partially denature the whey protein and adding a casein protein to the resulting solution. The temperature for the heat treatment is preferably 55 to 120°C, which is a temperature range sufficient to denature the non-denatured whey protein. The more preferable temperature range is 65 to 95°C. The solution is heated at these temperatures preferably for 1 to 120 minutes, and more preferably for 1 to 60 minutes. If the heating time is too short, the whey protein cannot be denatured and the degree of the hydrophobic property (hydrophobicity, Fl/mg-protein) which is hereinafter defined is decreased. If the heating time is extended beyond the above period, on the other hand, there is no added improvement in the shelf-life of the processed whey protein.

The degree of heat denaturation of the processed whey protein can be numerically evaluated by measuring the hydrophobicity. The hydrophobicity (Fl/mg-protein) of the processed whey protein is normally 50 Fl/mg-protein or more, and preferably 100 Fl/mg-protein or more. A whey protein gel with a good constitution cannot be obtained if the hydrophobicity is less than 50 Fl/mg-protein.

The hydrophobicity is expressed by the fluorescence intensity (FI) divided by the weight of whey protein (mg), wherein the fluorescence intensity is determined by diluting a whey protein solution to a concentration of 0.1 to 0.3 g protein/l, adding 8 mM of 1-anilinonaphthalene-8-sulfonic acid to the solution as a fluorescent probe, and measuring the fluorescence intensity at an exciting wavelength of 370 nm and a fluorescent wavelength of 470 nm using a fluorescent spectrophotometer. This method for the measurement of the degree of heat denaturation can be carried out according to the description in Japanese published unexamined patent application No. 64550/1993.

Next, a casein protein is added to a partially heat-denatured whey protein solution obtained according to the method described above. The amount of the casein protein to be added is 0.001 to 0.2 part by weight, preferably 0.005 to 0.1 part by weight, for 1 part by weight of the whey protein. Given as examples of the casein to be added are acid casein, rennet casein, sodium caseinate, potassium caseinate, magnesium caseinate, decomposition products of these caseins, casein micelles, and casein sub-micelles. The decomposition products of casein are the products obtained by decomposing a casein into a compound having a molecular weight of 1,000 or less. Either enzyme decomposition products or acid decomposition products can be used.

The presence of the casein protein added to a solution of partially heat-denatured whey protein can be confirmed by subjecting the product of the present invention to electrophoresis.

Detection of proteins is carried out by dipping the gel obtained after electrophoresis in a solution containing a dye substance which specifically combines with the proteins which have migrated to the gel by electrophoresis and then discoloring the solution. The isolation band (hereinafter simply called "band") of the migrated proteins can be confirmed as a dyed band. The dye used herein is a dye substance commonly used for dying electrophoresis gels and includes, for example, Coomassie Brilliant Blue, Amido black 10B, and First Green.

Although almost all proteins in the processed whey protein of the present invention are polymerized by the partially denaturation and are present as a soluble aggregate, the bands for non-polymerized  $\alpha$ -lactoalbumin or  $\beta$ -lactoglobulin, originally present in the whey protein, and the band for casein protein are detected as major components in the gel electrophoresis analysis. In the gel electrophoresis using a common acrylamide gel polymerized proteins such as the soluble aggregate cannot enter into the gel. Therefore, only the bands for non-polymerized  $\alpha$ -lactoalbumin,  $\beta$ -lactoglobulin, and the casein protein added can be detected when the processed whey protein of the present invention is subjected to electrophoresis by a conventional method. When a gel filtration analysis using high performance liquid chromatography is employed, partially heat-denatured whey proteins, non-polymerized  $\alpha$ -lactoalbumin and  $\beta$ -lactoglobulin can be detected.

In addition, according to the present invention a processed whey protein with improved shelf life can be obtained by adding a casein protein to a solution of a non-denatured whey protein and heating this solution to partially denature the whey protein, or by adding a casein protein to a solution of partially heat-denatured whey protein, which has been obtained by heating a whey protein solution, and adding a non-denatured whey protein to the resulting solution.

The presence of the non-denatured protein or casein protein in the processed whey protein is considered to improve the shelf life of the processed whey protein of the present invention. Because of the high hydrophobic characteristics of the protein molecules of the partially heat-denatured whey protein due to the soluble aggregate state of the whey proteins, the partially heat-denatured whey protein tends to be affected by the interactions between the whey protein molecules and other materials which are present together or the interactions between the partially heat-denatured whey protein molecules themselves. The casein protein added or the non-denatured whey protein is thought to suppress these interactions of the partially heat-denatured whey protein molecules.

The whey protein used in the present invention includes a cheese whey protein which is obtained as a by-product in a cheese manufacturing process, an acid whey protein produced by removing casein from milk by adding an acid, a whey protein produced by removing minerals and lactose from these whey proteins, and an isolated whey protein produced by recovering only precipitated whey protein fractions by adding ethanol. All of these can be prepared from milk and contain proteins such as  $\alpha$  -lactoalbumin and  $\beta$ -lactoglobulin. Among these whey proteins, particularly preferred are whey protein concentrate (WPC), whey protein isolate (WPI), and the like. The whey protein gels obtained from the WPC or WPI have a rigid, elastic, and smooth constitution.

When a casein protein is to be added previously to the non-denatured whey protein solution prepared under the above-mentioned conditions before the partially heat-denaturing is carried out, the casein protein is added to an aqueous solution of the non-denatured whey protein, and the mixture is blended to dissolution, following which the solution is heated to denature the whey protein.

The processed whey protein of the present invention obtained by the above-described process is soluble in water and form a gel if a salt is added into the solution at lower temperature or if heated after the addition of a salt. In addition, the processed whey protein stored for a long period of time can also produce a gel with a high water retention capacity, superior elasticity, and smooth constitution.

The processed whey protein obtained by the present invention exhibits a superior shelf life without an increase in the viscosity both during storage for a long period of time and thereafter. In addition, the gel obtained from the processed whey protein solution or powder which has been stored for a long period of time has a high water retention capacity, superior elasticity, a great gel strength, and a smooth constitution, and can be used in meat products such as

sausage and ham, desserts such as jelly, and fish meat products.

#### BRIEF DESCRIPTION OF THE DRAWING

Fig. 1 is an electrophoresis pattern showed by a chromatographic scanner of the processed whey protein obtained in Example 2 (Invention Product 6).

#### **DESCRIPTION OF PREFERRED EMBODIMENTS**

#### 10 Example 1

800 g of a commercially available whey protein concentrate (WPC Alacen 132<sup>™</sup>, protein content; 78.4%, ash content; 4.18%, pH 6.9) was dissolved in 9200 g of deionized water to produce a 8% WPC solution (protein concentration; 6.27%). This WPC solution was divided into five equal portions of 500 g each. Sodium caseinate (Sunlacto S-3(tm), a product of Taiyo Kagaku Co., Ltd.) or magnesium caseinate (a product of DMV Co.) were added with stirring in an amount such that the content of the casein protein is 0.001, 0.05, or 0.2 part by weight for 1 part by weight of whey protein. The mixtures were heated in a hot water bath while stirring to 78°C, at which temperature the mixtures were held for 30 minutes. Then, the mixtures were cooled to 5°C with ice water to obtain solutions (hereinafter designated as Invention Products 1-4). A solution without the addition of a casein protein was used as a Control Sample. Table 1 shows the amount of casein protein added for one part by weight of whey protein and the hydrophobicity of the solutions.

The solutions thus prepared were stored at 5°C for 3 weeks.

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TABLE 1

	Casein protein Amount (part by weight)	Hydrophobicity (FI/mg- protein)
Control Sample	•	102
	Na-caseinate	
Invention Product 1	0.001	104
Invention Product 2	0.05	103
Invention Product 3	0.2	95
	Mg-caseinate	
Invention Product 4	0.05	102

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#### Test Example 1

The properties of the solutions in the Invention Products 1 to 4 which were prepared in Example 1 and the Control Sample were confirmed immediately after preparation, one week, two weeks, and three weeks after the preparation.

Next, gels were prepared from the Invention Products 1-4 and the Control Sample. Sodium chloride was added to the Invention Products 1-4 and the Control Sample to a concentration of 1.2% at room temperature and the solutions were filled into cylindrical cells (diameter; 25 mm, height; 15 mm) and allowed to stand for 24 hours at 20°C, thereby obtaining gels. The gel strengths were measured using a Creep-Meter RE-33005™ (a product of Yamaden Co., Ltd.) with a circular disk plunger (diameter; 50 mm) at a compressibility ratio rate of 67% and a compression rate of 1 mm/sec. The gel strength (g) was expressed by the load at the maximum compression at 20°C.

Changes in the properties of the solutions during storage and the results of the gel strength measurement are shown in Table 2.

TABLE 2

		Immediately after preparation	After 1 week	After 2 weeks	After 3 weeks
Control Sample	Property of solution	Liquid	Liquid	Sol	Sol
	Gel strength (g)	1500	1450	<500	<500
Invention Product 1	Property of solution	Liquid	Liquid	Liquid	Liquid
	Gel strength (g)	1550	1560	1510	1510
Invention Product 2	Property of solution	Liquid	Liquid	Liquid	Liquid
	Gel strength (g)	1530	1520	1535	1490
Invention Product 3	Property of solution	Liquid	Liquid	Liquid	Liquid
	Gel strength (g)	1310	1290	1280	1300
Invention Product 4	Property of solution	Liquid	Liquid	Liquid	Liquid
	Gel strength (g)	1580	1580	1560	1530

All Invention Products 1-4 and the Control Sample were liquid immediately after the preparation. The viscosity of the Control Sample gradually increased during storage, and this sample became a highly viscous sol after two weeks. In contrast, the Invention Products 1-4 remained liquid after storage of three weeks with almost no increase in the viscosity.

With regard to the gel strength, all samples, the Invention Products 1-4 and the Control Sample, formed a solid gel from solutions immediately after preparation. After three weeks of storage, the Invention Products 1-4 exhibited almost no change in the gel strength as compared with that of immediately after the preparation, and the gels were solid. In contrast, the solution of the Control Sample partly gelled during storage. No homogeneous gel was obtained even if a salt was added.

The above results indicate that a solution of processed whey protein obtained by the addition of a casein protein to a partially heat-denatured whey protein exhibits an improved shelf life and that the gel obtained from such a solution of processed whey protein exhibits no decrease in the gel strength even if the solution has been stored for a long period.

#### Example 2

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600 g of a commercially available whey protein isolate (WPI, a product of Protose Separation Co., protein content; 78.4%, ash content; 4.18%) was dissolved in 9400 g of deionized water to produce a 6% WPI solution (protein concentration; 4.70%, pH 6.8). This WPI solution was heated in a hot water bath with stirring and held at 75°C for 20 minutes to obtain a partially heat-denatured WPI solution. The solution was cooled with ice water to 5°C. Then, the total 10 kg of the partially heat-denatured WPI solution was divided into five equal portions of 2 kg each. Sodium caseinate (Sunlacto S-3(tm), a product of Taiyo Kagaku Co., Ltd.) or magnesium caseinate (a product of DMV Co.) were added with stirring in an amount such that the content of the casein protein is 0.001, 0.05, or 0.2 part by weight for 1 part by weight of whey protein, thereby producing the products of the present invention (hereinafter designated as Invention Products 5-8). A solution without the addition of a casein protein was used as a Control Sample. Table 3 shows the amount of casein protein added for one part by weight of whey protein and the hydrophobicity of the solutions.

The solutions thus prepared were stored at 5°C for 3 weeks.

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TABLE 3

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	Casein protein Amount (part by weight)	Hydrophobicity (FI/mg- protein)
Control Sample	•	102
	Na-caseinate	
Invention Product 5	0.001	104
Invention Product 6	0.05	103
Invention Product 7	0.2	95
	Mg-caseinate	
Invention Product 8	0.05	102

#### 20 Test Example 2

The properties of the solutions in the Invention Products 5 to 8 which were prepared in Example 2 and the Control Sample were confirmed immediately after preparation, one week, two weeks, and three weeks after the preparation.

Next, gels were prepared from the Invention Products 5-8 and the Control Sample. 6 g (1.2%) of sodium chloride was added to the Invention Products 5-8 and the Control Sample at room temperature and the solutions were filled into cylindrical cells (diameter; 25 mm, height; 15 mm) and allowed to stand for 24 hours at 20°C, thereby obtaining gels. The gel strengths were measured using a Creep-Meter RE-33005(tm) (a product of Yamaden Co., Ltd.) with a circular disk plunger (diameter; 50 mm) at a compressibility ratio of 67% and a compression rate of 1 mm/sec. The gel strength (g) was expressed by the load at the maximum compression at 20°C.

Changes in the properties of the solutions and the results of the gel strength measurement are shown in Table 4.

TABLE 4

35			Immediately after preparation	After 1 week	After 2 weeks	After 3 weeks
	Control Sample	Property of solution	Liquid	Liquid	Sol	Sal
		Gel strength (g)	1600	1520	<500	<500
40	Invention Product 5	Property of solution	Liquid	Liquid	Liquid	Liquid
		Gel strength (g)	1610	1650	1580	1580
	Invention Product 6	Property of solution	Liquid	Liquid	Liquid	Liquid
		Gel strength (g)	1580	1550	1570	1550
45	Invention Product 7	Property of solution	Liquid	Liquid	Liquid	Liquid
		Gel strength (g)	1460	1510	1500	1480
	Invention Product 8	Property of solution	Liquid	Liquid	Liquid	Liquid
50		Gel strength (g)	1620	1640	1610	1620

All Invention Products 5-8 and the Control Sample were liquid immediately after the preparation. The viscosity of the Control Sample gradually increased during storage, and this sample became a highly viscous sol after two weeks. In contrast, the Invention Products 5-8 remained liquid after storage of three weeks with almost no increase in the viscosity.

With regard to the gel strength, all samples, the Invention Products 5-8 and the Control Sample, formed solid gels from solutions immediately after preparation. After three weeks of storage, the Invention Products 5-8 exhibited almost no change in the gel strength as compared with that of immediately after the preparation, and the gels were solid. In

contrast, the solution of the Control Sample partly gelled during storage. No homogeneous gel was obtained even if a salt was added.

The above results indicate that a solution of the processed whey protein obtained by the addition of a casein protein to a partially heat-denatured whey protein exhibits an improved shelf life and that the gel obtained from such a solution of processed whey protein exhibits no decrease in the gel strength even if the solution has been stored for a long period.

#### Example 3

7 kg of a whey protein isolate (WPI, Protose Separation Co., protein content; 89.5%, ash content; 2.3%) and 350 g of sodium caseinate were dissolved in 93 kg of deionized water to produce a 7.3% WPI solution (protein concentration; 6.5%, pH 6.90). This WPI solution was heated to 90°C in a hot water bath with stirring and held at this temperature for 15 minutes to obtain a partially heat-denatured WPI solution. Then, the total 100 kg of the solution was cooled to 50°C, and to 50 kg of the solution was added 3.5 kg of WPI powder and completely dissolved. The solution was spray dried using a vertical pressurespray drier to obtain Invention Product 9. The spray drier used a nozzle SX60-17(tm) manufactured by Spraying System Co., Ltd., and the spray drier was operated at a spray pressure of 175 kg/cm2, a hot air temperature of 168-176°C, and an exhaust gas temperature of 83°C.

Table 5 shows the amount of case in protein (g) and WPI powder (kg) added, the amount of powder recovered (kg), and the hydrophobicity.

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TABLE 5

	Invention Product 9
Na-caseinate (g)	350
WPI Powder (kg)	3.5
Recovered powder (kg)	4.85
Hydrophobicity (FI/mg-protein)	95

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#### Test Example 3

The Invention Product 9 obtained in Example 3 was stored for one month and three months at 20°C. Gels were prepared using the Invention Product 9 and a non-denatured whey protein (WPI) (Control Saample), immediately after preparation, one month, and three months after the preparation. WPI was dissolved in 166 g of deionized water to a concentration of 15%. Then, 4 g of sodium chloride was added to the solution. This solution was filled into a polyvinyl chloride tube (diameter 3 cm) of which the one end was tied off with a string. After closing another end by tying off with a string, the solution was allowed to stand for 15 minutes, then the tube was placed in a hot water bath at a temperature of 75°C for 45 minutes to gelatinise the solution. The tube was removed from the hot water bath, cooled in a stream of water for 30 minutes, and allowed to stand at 5°C overnight, followed by measurement of the gel strength. The gel strength was measured in the same manner as in the Test Example 1 using a Creep-Meter RE-33005(tm) (a product of Yamaden Co., Ltd.).

The water dissociation was determined by preparing a gel in the same manner as in the measurement of the gel strength, placing the gel with a diameter of 3 cm and a length of 3 cm on a filter paper with a diameter of 12.5 cm (No. 50 Whatman<sup>TM</sup>), and measuring the area [(length of the long axis )x(length of the short axis)] of filter paper into which water was absorbed in 10 minutes by approximation.

Table 6 shows changes in the gel strength and the water dissociation, measured immediately after preparation, and one month and three months after the preparation.

TABLE 6

Invention Product 9	Control Sample
4720	4880
4680	4530
4700	4570
24.4	50.3
25.8	48.8
26.0	48.0
	4720 4680 4700 24.4 25.8

Both the Invention Product 9 and the Control Sample showed approximately the same gel strength immediately after preparation. After one month and three months storage, the gel strength of the Invention Product 9 was maintained at almost the same level as that immediately after preparation without any decrease. The gel obtained was smooth and elastic as compared with the gel obtained from the Control Sample. In addition, the powder of the Invention Product 9 exhibited superior solubility.

The Invention Product 9 showed a low water dissociation after storage for one month or longer. The water dissociation of the Invention Product 9 was excellent as compared with that of the Control Sample.

#### Test Example 4

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The solution of the Invention Product 6 obtained in Example 2 was mixed with an SDS electrophoresis buffer solution (0.125 M Tris-glycin buffer; pH 6.8, 1% SDS, 40% glycerol) to prepare a sample for electrophoresis. After electrophoresis using SDS-polyacrylamide gel under unreduced conditions, proteins were dyed with Coomassie Brilliant Blue G-250™ (a product of Sigma Co.), followed by discoloration using 10% methanol and 7.5% aqueous solution of acetic acid. The detected protein bands were subjected to a chromatographic scanner CS-930™ (a product of Shimadzu Corporation) to examine the electrophoresis pattern of the Invention Product.

The results are shown in Fig. 1.

As shown in Figure 1, the bands of casein were clearly detected in the electrophoresis of the Invention Product 6, confirming the addition of the casein.

#### 40 Example 4

## (Preparation of dessert)

1 kg of a solution of the Invention Product 2 prepared in Example 1 was heated to 45°C, and 0.8 kg of skimmed milk, 1.0 kg of granulated sugar, and 0.01 kg of almond flavor were mixed. The mixture was allowed to stand for 4 hours to obtain a gel (at this instance, salts contained in the mixture were mainly calcium ion from the skimmed milk with an estimated concentration of 0.5%). The resulting gel was cut into 1cm x 1cm 1 x 1cm squares and mixed with a syrup or syrup-dipped fruit to prepare a dessert.

The gel strength measured before dipping in syrup confirmed that the gel was rigid with a gel strength of about 800g.

Upon prelibation, the dessert was found to have an elastic, smooth texture and the taste and external appearance were excellent.

The gel strength was measured in the same manner as in the Test Example 1 using the Creep meter.

A gel was prepared in the same manner using a solution of non-denatured whey protein instead of the Invention 55 Product 2, but the solution did not gel.

#### Example 5

#### (Preparation of low-fat sausage)

6 kg of lean pork was ground in a meet grinder and all the meat was put into a silent cutter. 133 g of WPI powder of the Invention Product 9 prepared in Example 3 was added. Then, 1.2 kg of ice water, 150 g of salt, 30 g of phosphate, 1.6 g of nitrite, 10 g of glutamate, 11 g of sugar, 6 g of a flavor, and 320 g of potato starch were added and mixed, while cutting at high speed, to obtain a paste. The paste was filled into a tube and steamed so as to heat the center to 70°C, then cooled to obtain a law-fat sausage. Another low-fat sausage was prepared in the same manner using non-heated WPI powder instead of the Invention Product 9 (Comparative Example 2)

The yields of the resulting low-fat sausages were measured and sensory evaluation was carried out. The results are shown in Table 7.

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#### TABLE 7

	Yield	Sensory evaluation
Low-fat sausage of the present invention	93.1	Excellently chewed and swallowed.
Cpmparative Example 2	89.0	The texture was dry with no juicy sensation.

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The sausage prepared from the Invention Product 9 was superior to that of the Comparative Example 2 and a high yield was obtained. In the sensory evaluation, the sausage of Comparative Example 2 exhibited no juicy sensation, the meat was dry, and a liquid flowed from the incision, whereas the low-fat sausage prepared from the Invention Product 9 was easily chewed and swallowed.

#### INDUSTRIAL APPLICABILITY

The processed whey protein of the present invention has an improved shelf life both in a solution and in a form of a powder. The gel which is obtained from the processed whey protein exhibits improved water retention capacity and elasticity, and has a smooth constitution. The processed whey protein of the present invention can be used as a gelling agent, water preserving agent, viscosity increasing agent, and the like.

# 35 Claims

- A processed whey protein with an increased shelf life comprising a partially heat-denatured whey protein and a casein protein.
- The processed whey protein according to claim 1, wherein a partially heat-denatured whey protein is prepared by providing an aqueous solution of the non-denatured whey protein with a protein concentration of 0.5 to 15 wt% and pH 6-8, and heating this solution at 55 to 120°C for 1 to 120 minutes.
- 3. The processed whey protein according to claim 1 or 2, wherein the casein protein is at least one protein selected from the group consisting of acid casein, rennet casein, sodium caseinate, potassium caseinate, magnesium caseinate, and decomposition products of these caseins.
  - 4. The processed whey protein according to any one of claims 1 to 3, containing a casein protein in an amount of 0.001 to 0.2 part by weight for 1 part by weight of a partially heat-denatured whey protein.

- The processed whey protein according to any one of claims 1 to 4, wherein the processed whey protein has a hydrophobicity of 50 FI/mg-protein or greater.
- 6. A powder exhibiting excellent gelling properties when dissolved in water, which is prepared by drying a solution containing a partially heat-denatured whey protein and a casein protein.
  - A process for preparing a processed whey protein comprising adding a casein protein to an aqueous solution of a non-denatured whey protein and heating the solution to partially heat-denature the non-denatured whey protein.

- 8. A process for preparing a processed whey protein comprising preparing an aqueous solution of a partially heatdenatured whey protein by heating an aqueous solution of a non-denatured whey protein and adding a casein protein to this solution.
- 9. The process for preparing a processed whey protein according to claim 7 or claim 8, wherein the aqueous solution of the non-denatured whey protein has a protein concentration of 0.5 to 15 wt% and pH 6-8.

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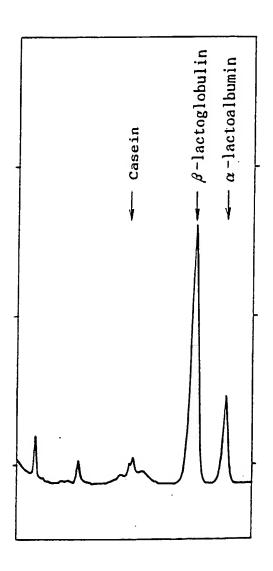
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- 10. The process for preparing a processed whey protein according to claim 7 or claim 8, wherein the solution is heated at 55 to 120°C for 1 to 120 minutes.
- 11. The process for preparing a processed whey protein according to any one of claims 7 to 10, wherein a casein protein in an amount of 0.001 to 0.2 part by weight is used for 1 part by weight of a partially heat-denatured whey protein

Fig. 1



# INTERNATIONAL SEARCH REPORT International application No. PCT/JP95/02553 CLASSIFICATION OF SUBJECT MATTER Int. Cl<sup>6</sup> A23J3/08 According to International Patent Classification (IPC) or to both national classification and IPC Minimum documentation searched (classification system followed by classification symbols) Int. C16 A23J3/08 Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) WPI, WPI/L C. DOCUMENTS CONSIDERED TO BE RELEVANT Category\* Citation of document, with indication, where appropriate, of the relevant passages Relevant to claim No. Х JP, 5-260898, A (Snow Brand Milk Products Co., 1 - 11 Ltd.), October 12, 1993 (12. 10. 93) (Family: none) JP, 5-64550, A (Snow Brand Milk Products Co., Α 1 - 11Ltd.) March 19, 1993 (19. 03. 93) & US, 5217741, A Α JP, 4-229145, A (Quest International B.V.), 1 - 11 August 18, 1992 (18. 08. 92) & EP, 485663, A1 & CA, 2038134, A & AU, 9172827, A & DE, 69004096, E & ES, 2045770, T3 Further documents are listed in the continuation of Box C. See patent family annex. later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "X" document of particular relevance; the claimed invention cannot be considered acovel or cannot be considered to involve an inventive "E" earlier document but published on or after the international filing date document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) considered novel or cannot be considered step when the document is taken alone document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "O" document referring to an oral disclosure, use, exhibition or other document published prior to the international filing date but later than the priority date claimed "&" document member of the same patent family Date of the actual completion of the international search Date of mailing of the international search report March 14, 1996 (14. 03. 96) April 2, 1996 (02. 04. 96) Name and mailing address of the ISA/ Authorized officer

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